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The Occurrence of Trematode Ova, *Hapalotrema constrictum* (Leared), in Fibro-epithelial tumors of the Marine Turtle, *Chelonia mydas* (Linnaeus).

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(Plates I-V).

In an earlier paper a description was given of a cutaneous disease occurring in large marine turtles, *Chelonia mydas* (Linnaeus), captured in waters south of Key West (Smith & Coates, 1938). This disease is characterized by multiple fibro-epithelial tumors varying in size, occupying chiefly the cutaneous region of the neck, axillae, groins, eyelids, the conjunctivae and the cornea (Figs. 1, 2). The tumors followed two types: a papillary or coral-like growth with hyperplasia of keratinizing epithelium supported by a firm fibrous core; and a smooth round or oval fibrous form of tumor composed of dense and frequently hyalinized connective tissue covered externally by slightly thickened epithelium. At times both types of growth co-existed, thus forming confluent masses in which both forms of growth are distinguishable (Fig. 3). When tumors arose from pigmented skin, they were of a dark gray or black color, due to the large number of perivascular melanophores in the substance of the growth.

Histologically the fibro-epithelial tumors of *Chelonia mydas* seem to be of benign character. Epithelium of papillomas may show extensive downgrowth into the stroma with "pearl" formation. In one tumor, which was taken from the lower eyelid, an irregular adenomatous change occurred, somewhat suggestive of early malignancy (Fig. 4).

In our earlier observations, the possibility of a virus origin for these tumors was discussed. It was thought that larger parasites or their ova were a negligible factor in the disease. However, the study of larger amount of material, consisting of about 250 tumors, indicates that in more than half of the tumors parasitic ova can be detected readily with the aid of a dissecting microscope in thin slices of gross tumor tissue or in prepared microscopic sections. The ova may exist at the bases or in other parts of the stroma of both small tumors (3 mm.) as well as in larger growths (Figs. 5, 6, 7). They are present in both the papillomatous or fibromatous type of growth. Ova may be widely scattered in the fibrous stroma, or arranged in collections (Fig. 9). Frequently they occupy tissue spaces which resemble small venules or lymphatics. There is at times

found a mild inflammatory reaction about the ova or in adjacent tissue composed of lymphocytes, leukocytes and fibroblasts. Occasionally there are eosinophilic cells in the tumor. Parasitic ova lying either in the region of hyperplastic surface epithelium or in the deeper fibrous stroma of the growth usually exhibit epithelioid cells distributed along the outer surface of the chitinous membrane of the egg (Fig. 8). Such epithelioid cells may be fused into a syncytium or form multinucleated giant cells. At times, ova seem to elicit no cellular response whatever. Normal skin attached to the tumors has appeared to be free of ova.

Ova are yellow in color and consist of a body with two polar filaments. The covering is a firm chitinous-like membrane. The cellular arrangement of the egg enclosed within the membrane suggests often the first cleavage stage of larvae (miracidium). Ova removed from tumor tissue fixed in 10% formalin (Figs. 9, 10) show a length (including filaments) of 260-310 microns and a width (in middle of miracidium) of 30-40 microns.

It is thought that the ova found associated with the turtle fibro-epithelial tumors are identical with or related to those belonging to a trematode or blood fluke described by Looss (1902) and referred to as *Hapalotrema constrictum* (Leared). Both the morphology of the eggs and their measurements correspond to descriptions given for *Hapalotrema constrictum*. This digenetic trematode belonging to the family Spirorchidae was found by Looss (1898-9) in the blood and visceral tissues of two species of turtles, *Thalassochellys cortica* and *Chelonia mydas* caught along the coast of Egypt.¹

It is probable that ova are deposited in pre-existing vascular tumor tissue by the migrating blood fluke, and remain there without affecting the subsequent course of the growth. The localization of ova in the stroma and in the venous or lymphatic spaces of turtle tumors recalls a similar localization of ova in the tissues of the human bladder in bilharziosis. Such an infection of the human bladder, as is well known, may result in papillomatous and malignant changes. Phylogenetically, Hapalotrema constrictum of turtles and Schistosoma haematobium responsible for bilharziosis are closely related.

During the past year, two smaller specimens of *Chelonia mydas* (150 lbs.), caught in Biscayne Bay, Florida, have been kept under observation for a period of about three months in the large salt water tanks of the New York Aquarium.² It is noteworthy that at autopsy no evidence of ova were found in heart, lungs, liver, spleen or kidneys of these two turtles, nor were there ova in any of the numerous cutaneous tumors or in those of the eyelid and cornea, although a great many microscopic preparations were examined.

These last findings indicate, therefore, that the parasitic ova of such turtle tumors are probably not of primary importance as a factor in the etiology of the disease. The blood fluke itself has not been found in the tumors examined.

SUMMARY.

The presence of parasitic ova of a blood fluke, *Hapalotrema constrictum* (Leared), has been noted in many of the growths which occur in the marine turtle, *Chelonia mydas*. This neoplastic disease in the turtle is characterized by multiple cutaneous growths and may at times involve the eyelids and cornea. The parasitic ova, as such, probably do not act as the immediate cause of the disease.

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² These two specimens were received through the kindness of Dr. Thomas Otto of Miami, Florida. Photographs of the live specimens were made by Mr. S. C. Dunton of the New York Aquarium.

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EXPLANATION OF THE PLATES.

PLATE I.

Figs. 1 & 2. Chelonia mydas (Linnaeus), weight 150 lbs., showing distribution of fibro-epithelial tumors.

PLATE II.

- Fig. 3. Tumor mass composed of confluent papillomas (P) and fibromas (F).
- Fig. 4. Papillomatous tissue from an eyelid. Extreme adenomatous hyperplasia. ×30.

PLATE III.

Figs. 5 & 6. Cross-section of ova (X) located at base of a small papilloma. $\times 75$ and $\times 150$.

PLATE IV.

- Fig. 7. Fragments of ova (X) located in the stroma of a papilloma. ×80.
- Fig. 8. Cross-section of parasitic egg (X) surrounded by epithelioid cells (E). ×250.

PLATE V.

- Fig. 9. Collection of ova lying in a thin slice of tumor tissue, photographed by direct illumination. Actual measurements given in text.
- Fig. 10. Teased preparation of single parasitic egg, of *Hapalotrema constrictum* shrunken after fixation in 10% formalin. For measurements see text.